Health Risk Assessment of Nitrite and Nitrate in the Drinking Water in Mashhad, Iran

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Introduction

In the past decade, the quality of water resources has been threatened, especially due to population growth, urbanization, and agricultural activities, as well as the unprocessed disposal of wastewater into the environment (1-4). According to recent studies,
approximately 80% of diseases and more than 33% of deaths in developing countries are caused by the consumption of contaminated water (5). Disregard of environmental standards, especially in metropolises, has led to the more complications in relation to the pollution of water resources.

Among various pollutants, nitrate and nitrite are of paramount importance (6). Recently, the concentration of nitrate has been reported to increase in surface waters and groundwater across the world (7). Nitrate is a common pollutant in the aquifers across the country due to the anthropogenic activities in underground water (8).

In the human body, nitrates and nitrite could lead to the formation of carcinogenic compounds. The International Agency for Research on Cancer (IARC) has classified nitrate as a carcinogenic agent (9). In infants aged less than six months, nitrate could lead to the development of methemoglobinemia, also known as the blue baby syndrome (10), which may even cause neonatal mortality (11). In adults, nitrate may cause cancer and fetal defects (12). The epidemiological studies conducted by Grosse et al. at the IARC in the United States have indicated that the consumption of nitrates and nitrites is associated with the increased prevalence of some cancers of the stomach and esophagus, as well as other cancers (13). Moreover, several epidemiological studies of various human populations have shown a correlation between gastric cancer and nitrate in drinking water (10).

Drinking water is one of the sources of nitrate entry into the human body and the subsequent adverse health effects. In research, nitrate has attracted great attention as a common, extensive pollutant of groundwater within the past decade (2, 11, 14, 15). According to the studies performed in Shiraz (Iran), the nitrate concentration in 11% of drinking water samples was above the standard levels (16). Additionally, nitrate concentration was reported to be 1.8-82.2 mg/l in Gonabab (Iran) and 5.5-84.3 mg/l in Bajestan (Iran) in 2018 (17).

Risk assessment involves the identification and quantification of the risks associated with the use of chemical substances through considering their possible effects on the consumers of these chemicals via various exposure pathways. Risk assessment was introduced by the United States Environmental Protection Agency in 1983 (18). The main objectives of risk assessment include identifying the hazards associated with each chemical, assessing the degree of exposure to harmful or toxic chemicals, evaluating the hazards associated with harmful or toxic chemicals, and determining the possible adverse effects of exposure to these chemicals (19).

Mashhad is the second metropolitan city in Iran with the population of three million. In this city, several factors influence the water supply, leading to the increased concentrations of nitrate and nitrite. Some of these factors are sewage disposal into absorbent wells, non-standard disposal of municipal solid waste, and the inappropriate location of the industrial estate in the upstream of water resources (20). There are limited studies regarding the risk assessment and exposure of individuals to nitrate and nitrite contamination through drinking water in Iran (17, 18). In addition, the sources and effects of nitrate and nitrite in Mashhad remain unknown, and there have been few advanced surveys on the nitrate and nitrite contamination induced by the spatial and temporal changes in Mashhad. This paper represents the first study regarding the exposure assessment of nitrate and nitrite in Mashhad.

Given the importance of the Mashhad as the religious capital of Iran, as well as the booming tourism and industrial centrality of this metropolitan, it is essential to conduct such investigations in order to control the quality of drinking water for human health preservation. The present study aimed to determine the concentrations of nitrate and nitrite in the drinking water of Mashhad distribution network and examine the possible health risks.

Material and methods

Mashhad is the second metropolitan city in Iran, located in the center of Khorasan Razavi province at the longitude of 58° 20'-60° 8’ and latitude of 35° 40'-36° 3’ (Figure 1). This city covers a total area of 313 square kilometers and is located in the catchment area between the mountains of Hezarmasjed and Binaloud (20).
In this cross-sectional study, 36 stations were selected from five zones (north, south, east, west, and center) in Mashhad based on population density, easy access, and land use (Figure 1, Table 1). The stations were sampled twice in spring and summer during March-September 2017, and 72 water samples were collected. The location of the sampling points was determined using a GPS receiver (GPS Garmin Montana, model: 5S, USA). The collected samples were transferred to the laboratory at the temperature of 4°C and analyzed immediately.

Table 1. Address and Coordinates of Sampling Stations

<table>
<thead>
<tr>
<th>Sampling Station</th>
<th>Address</th>
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<td>721779</td>
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<td>Mosalla 14</td>
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<td>22 Bahman 16</td>
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Figure 1. Location of Sampling Stations and Zones in Mashhad, Iran
Sample analysis was based on the standard method 4500 and performed using a spectrophotometer (model: HACH 6000-DR, USA)(21). Nitrate and nitrite concentrations were measured in accordance with the 355 and 371 spectrophotometer programs at the wavelength of 500 and 507 nanometers using NitraVer®5 and NitriVer®3 reagents and compared to the standard levels. Nitrate and nitrite concentrations in each sample were measured three times, and the mean concentrations were determined. According to the Iranian standards for drinking water (22) and the World Health Organization (WHO) guidelines (23), the maximum acceptable concentrations (MAC) of nitrate and nitrite are 50 and 3 mg/l, respectively.

At the next stage, the health risks associated with nitrate and nitrite were evaluated in children and adults using the recommended human health risk assessment model of the USEPA (24, 25). The non-carcinogenic health risk assessment of nitrate and nitrite was obtained based on equations 1 and 2, as follows:

\[
ADD = \frac{C \times IRd \times EF \times ED}{BW \times AT} \quad \text{Equation (1)}
\]

\[
THQ = \frac{ADD}{RfD} \quad \text{Equation (2)}
\]

where \(ADD\) is the average daily dose of the elements through ingestion pathways (mg/kg/day), \(C\) represents the concentration of the elements (mg/l), \(IRd\) shows the daily ingestion rate (l/day; average consumption rate: 1.8 and 2 liters per day in children and adults, respectively), \(BW\) denotes the body weight of the target groups (31.22 and 70 kilograms, respectively), \(ED\) is the duration of human exposure (six and 70 years in children and adults, respectively), \(EF\) shows the exposure frequency (365 days/year), \(AT\) represents the average time of human exposure (\(ED \times 365\) days), and \(RfD\) denotes the oral reference dose (mg/kg/day), indicating the continuous, daily exposure of the human population over a lifetime without the appreciable risk of deleterious effects.

It is notable that the reference dose for nitrate and nitrite was 1.6 and 0.1 mg/kg/d, respectively. With the hazard quotient (HQ) of less than one, the population is assumed to be safe and exposed to no health risks (24).

Data analysis was performed in SPSS (Chicago, IL) and Excel (Microsoft Office 2007) using bivariate tests to determine the significant variation of nitrate and nitrite concentrations in various stations and seasons.

**Results**

The statistical descriptions of nitrate and nitrite values are presented in Table 2. The analysis of drinking water in Mashhad demonstrated significant differences between the distribution of nitrate and nitrite at various stations \((P<0.05, \text{ sig.} = 0.001)\), while no significant difference was observed between the concentrations of these elements in various seasons \((P>0.05, \text{ nitrate sig.} = 0.056, \text{ nitrite sig.} = 0.072)\).

Nitrate concentration was within the range of 5.6-49.8 mg/l with the mean concentration of 16.63±10.88 mg/l in Mashhad city (Figure 2).
Nitrite concentration was within the range of 0.009-0.039 mg/l with the mean concentration of 0.02±0.01 mg/l (Figure 3). The average nitrate concentration was estimated at 9.6, 26.01, 18.77, 13.66, and 16.5 mg/l, while the average nitrite concentration was estimated at 0.015, 0.027, 0.02, 0.019, and 0.023 mg/l in the northern, southern, eastern, western, and central zones, respectively.

Figure 2. Comparison of Nitrate Concentration with Maximum Recommended Concentration

Figure 3. Comparison of Nitrite Concentration with Maximum Recommended Concentration

According to the findings, the concentrations of nitrate and nitrite in all the stations reached lower levels than the maximum contaminant level recommended in the national standards of drinking water in Iran and WHO guidelines (nitrate: 50 mg/l, nitrite: 3 mg/l). The average
daily dose (ADD) of nitrate through ingestion pathways was within the range of 2.87-0.32 mg/kg/day in children, while it varied within the range of 1.423-0.16 mg/kg/day in adults. The average ADD of nitrite changed from 0.0022 to 0.0005 mg/kg/day in children and from 0.0011 to 0.0003 mg/kg/day in adults (Table 2).

A summary of the HQ values for nitrate and nitrite in drinking water through ingestion in adults and children is depicted in Figures 4 and 5. As can be seen, the HQ values through ingestion exposure did not exceed the threshold of the HQ for adults (<1), while the value was higher than one in children in some stations, including stations 11, 13, 14 (southern zone), 21 (eastern zone), 28 (western zone), and 33 (central zone). The HQ of nitrate was within the range of 0.2-1.795 in children and 0.1-0.889 in adults. Additionally, the range of HQ for nitrite was 0.0003-0.0014 in children and 0.002-0.0007 in adults (Table 2). It is also notable that the HQ in children was twice higher than the value obtained in adults in all the stations.
Discussion

According to the results of the present study, the nitrate and nitrite concentrations in drinking water had insignificant variations on the temporal scale, which could be due to the insignificant changes in the atmospheric deposition and pollution load of the anthropogenic sources in two seasons. Several studies have indicated that these factors could cause temporal variations in the dilution, mobility, chemical properties, bioavailability, and enrichment of nitrate and nitrite during two seasons (26).

In the current research, the nitrate and nitrite concentrations in drinking water showed significant variations on the spatial scale, which could be due to the differential derivation of these pollutants from various water supplies and purification systems. Evidently, the drinking water in Mashhad is derived from various sources, including Dosti dam (southern and eastern zones), water wells, and groundwater (northern and western zones), and both these sources at the same time (central zone) (27).

In the present study, nitrate concentration was within the range of 5.6-49.8, and the
maximum nitrate concentration was observed in the southern and eastern areas of Mashhad, particularly in stations 11 (Hor 25), 13 (17-Shahriyar), and 14 (Chaman 23). This finding was attributed to the nitrate contamination of the water supply from which the drinking water in these stations is derived. Dosti dam (located at Turkmenistan and Iran border) is the major source of drinking water in the southern and eastern areas of Mashhad (28, 29), and the high concentration of nitrate could be attributed to the water quality in this dam. A growing number of studies have reported that factors such as successive droughts, thermal layering, severe evaporation from the water surface, chemical reaction of the reservoir floor with dam water, and the construction of Salma dam at the top of the dam in Afghanistan, have affected the water quality in Dosti dam (28, 29).

According to the literature, the nitrate load in Dosti dam in spring and summer is derived not only from atmospheric deposition and situ nitrification, but also from other sources, such as agricultural activities (30). This might affect the quality of drinking water in the southern and eastern areas of Mashhad compared to the other regions in this city. In this regard, our findings are inconsistent with the results obtained by Latifi et al. (2005), which indicated that the high concentration of nitrate was distributed along the southern and central regions in Mashhad (31). However, the nitrate concentration in the present study was lower (16.63±10.88 mg/l) compared to the concentration reported in the mentioned study (23.1±11.15 mg/l) (29). Therefore, it could be inferred that the concentration of nitrate has decreased within the past decade, which could be due to the improvement of purification systems, reconstruction of the distribution network in some areas, and closing of the sewage wells with the high dispersion of nitrate (32).

In the current research, the concentrations of nitrate and nitrite in all the stations were lower compared to the maximum contaminant level recommended by the national standards of drinking water in Iran and WHO guidelines. Our findings in this regard are consistent with the domestic studies conducted by Panahi et al. in Tehran (33), Nan Baksh et al. in Orumiyeh (34), and Marboti et al. in Khuzestan (35), while inconsistent with the results obtained by Ziarati et al. in Gilan (36), Jafari (37), Ismaili in Isfahan (38), Khazaei et al. in Sistan and Baluchestan (28), and Moghadasi et al. in Arak (29). However, some studies in Iran have reported nitrate concentrations to be higher than the standard level. In this regard, the studies conducted in Shiraz have demonstrated that nitrate concentration in 11% of drinking water samples was higher than the standard level (16). In addition, nitrate concentration was reported to be 1.8-82.2 mg/l in Gonabad (Iran) and 5.5-84.3 mg/l in Bajestan (Iran) in 2018 (17).

According to the current research, the health risks for adults through ingestion exposure were lower than the recommended values by the Environmental Protection Agency (HQ<1). However, the health risks for children was higher than the recommended values by the Environmental Protection Agency (HQ>1) in stations 11, 13, 14 (southern region), 21 (eastern region), 28 (western region), and 33 (central region). Therefore, it could be inferred that there is no non-carcinogenic threat from nitrate and nitrite through the daily intake of drinking water in the adult population in Mashhad, while non-carcinogenic risks were observed in children in some of the stations in Mashhad. It is also notable that the estimated risks in children were higher compared to adults, which indicated that children were more susceptible to the health risks associated with nitrate and nitrite contamination. This finding is in line with the majority of the studies in this regard, reporting that children are affected by nitrate contamination more significantly compared to adults since they consume more water per kilogram of their body weight (30). Moreover, the immune, digestive, reproductive, and nervous systems of children are still growing, and exposure to toxic substances could cause irreversible damages to these systems (39).

In a similar study, Su et al. investigated the groundwater in the northern provinces of northern China and the associated non-carcinogenic health risks, reporting that the health risks were more significant in children compared to adults (40). In another research, Chen et al. (2017) examined the health risks of nitrate in the underground water in northwestern China in infants, children, and
adults. According to the findings, the risk index for infants and children was higher than adults, and it is possible that nitrates damage these two age groups more significantly (41). In addition, Ghasemi et al. (2018) investigated the drinking water in the rural areas of Bajestan and Gonabad (Iran) in terms of health risks, reporting that infants were more significantly exposed to the health risks of water contaminants compared to children and adults (17). Similarly, Rezaei et al. (2017) claimed that children were more significantly exposed to the health risks associated with elements such as nitrates in drinking water compared to other populations (42).

Conclusion

According to the results, nitrate and nitrite concentrations were lower than the standard values in all the studied stations. Furthermore, the results of health risk assessment indicated that the health risks exceeded the recommended levels for children in some areas of Mashhad city, which could pose severe threats to their health. In this study, health risks were determined based on average ingestion and average concentration, as well as the uptake and intake rates over the highest exposure time. This possibility could be considered a factor of uncertainty in risk estimations.

The health risk assessment in the present study was only focused on the concentrations of nitrate and nitrite, while drinking water contains other elements as well (e.g., heavy metals and organic compounds). Therefore, the level of risks associated with drinking water in Mashhad may be higher than the calculated values in this research. Based on this finding, further efforts are required to decrease the nitrate concentration in drinking water in Mashhad. Some suggested measures in this regard include the control of nitrate discharge and specific regulations to halt the release of contaminants into water supplies (e.g., water wells, groundwater, and Dosti dam). Furthermore, effective wastewater treatment methods must be applied in order to reduce the concentration of nitrate in drinking water.

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